partner, the analyte binding partner being present in an amount from 10^9 to 10^{12} molecules per each sorbent zone with a diameter from 60 μm to 500 μm.

36. (New) The analyte binding array of claim 35, wherein the analyte binding partner forms a multi-layer matrix in the sorbent zone.

REMARKS:

Claims 1, 23 and 26 are amended; marked up versions of the amended claims are attached hereto pursuant to 37 C.F.R. § 1.121(c)(ii). The support for the amendments can be found on page 19, lines 16-24, of the specification. New claims 29-36 are added. The support for claims 29 and 34 can be found on page 13, lines 27-31, and page 17, lines 25-30. The support for claims 30-33, 35 and 36 can be found on page 11, lines 4-10 and page 19, lines 16-33, of the specification. No new matter has been introduced. Claims 1-36 are pending in the application. Reexamination and reconsideration of the application, as amended, are respectfully requested.

Claims 1-4, 13-19, 21 and 23-28 are rejected under 35 U.S.C. § 103(a) as being unpatentable over Ekins et al., EP 304,202 (the '202 patent) in view of Ekins et al., *Journal of Clinical Immunology*. (the Immunology reference). The grounds of the rejections in the outstanding Office Action are substantially the same as in the previous Office Action. This rejection is respectfully traversed.

The Examiner found the applicants' arguments in the preliminary amendment filed on May 13, 2002 not to be persuasive because the rejected claims do not recite the limitations that form the basis for the applicants' arguments (page 4 of the Office Action). In particular, the Examiner noted that the rejected claims do not recite that blocking of the lower layers of the analyte binding partner by the top layers does not occur. The Examiner also stated that other features upon which applicants relied in the arguments, such as "immobilized binding partner molecules extending up to 200 nm vertically from the surface of the film, the use of photo-

linking techniques and printing antibody at concentrations 1000 times greater then the 1 μm solutions" were not recited in the rejected claims. The Examiner made similar comments with respect to the arguments presented in the declaration of Dr. Silzel (Declaration) (pages 10-11 of the Office Action).

In response, applicants amended independent claims 1, 23 and 26 by adding a limitation "a microscopic sorbent zone comprises a multi-layer matrix of an analyte binding partner." The added term "multi-layer matrix" is supported by the description on page 19, lines 16-26, of the instant specification as read in view of the knowledge of those skilled in the art. Specifically, the specification provides an example of a sorbent zone with immobilized avidin molecules. The sorbent zone has "an irregular topology extending up to 200 nm vertically from the surface of the film [substrate]," wherein "each avidin molecule occupies 6nm^3 in the dry matrix."

The specification provides a sufficient written description of the term "multi-layer matrix" although the terminology used in the claim differs from that in the specification. In its recent decision in All Dental Prodx, LLC v. Advantage Dental Products, Inc. (002-1107; decided October 25, 2002), the Federal Circuit observed that "the failure of the specification to specifically mention a limitation that later appears in the claims is not a fatal one when one skilled in the art would recognize upon reading the specification that the new language reflects what the specification shows has been invented." Thus, based on the description on page 19 of the specification and a general knowledge of molecular structure, those skilled in the art would have understood that applicants, at the time the application was filed, had possession of the "multi-layer matrix" of analyte binding partner.

The '202 patent taken in view of the Immunology reference does not make the amended claim obvious. Amended claims 1, 23 and 26 require a multi-layer matrix of an analyte binding partner that allows substantial depletion of the analyte from the sample and its concentration on the microscopic sorbent zone. The '202 patent and the Immunology reference have no teaching whatsoever of a multi-layer matrix of an analyte binding partner immobilized in the sorbent zone, much less of such

multi-layer matrix that is capable of substantial depletion of the analyte from the sample.

The '202 patent and the Immunology reference do not inherently teach a multi-layered matrix of the analyte binding partner because they utilize significantly lower concentrations of the binding partner. Example 1 (page 19, lines 21-33) of the present invention describes a biotin/avidin binding system. In that system, 10^9 avidin molecules were bound per spot with a diameter of 100 μm ("The printed avidin spots ... were found to have a binding capacity on the order of 10^9 dye-biotin molecules per spot, ... assuming one binding site per avidin molecule"). The area of such spots can be easily calculated ($0.785 \times D^2$) and equals about 0.01 mm^2. Therefore, in the present invention, 10^9 molecules were bound per 0.01 mm^2 or 10^{11} molecules per mm^2. On the other hand, Example 1 of the '202 patent teaches depositing $5x10^9$ molecules of antibody per mm^2 of the spot area. Therefore, the concentration of the analyte binding partner in the sorbent zone of the present invention is almost two orders of magnitude higher than that of the cited art. Because the spots of the '202 patent have a much lower analyte-binding capacity and, thus, cannot concentrate analyte, a weaker fluorescent signal is generated. In fact, Figure 1 of the present invention demonstrates that the signal of the ambient assays of the cited references is from 60 to 100 times weaker than the signal produced in the assay of the present invention.

The cited references, either alone or in combination, do not make the multi-layer matrix of an analyte binding partner obvious. As explained in the previously filed preliminary amendment, the concept of a substantial analyte depletion from the solution and the concentration of the analyte onto multi-layered microscopic sorbent zones is not obvious in view of the cited references because it is based on an unexpected discovery. The present invention is based on a discovery that the binding capacity of a conventional microplate well may be concentrated onto an area of a sorbent zone that is 100 times smaller than that of the microplate well without a significant loss of analyte-depletion capability. This result is achieved by immobilizing relatively large quantities of the binding partner on each sorbent zone,

thereby forming a multi-layered matrix of the binding partner. As explained above, the cited references teach significantly lower levels of the binding partner and, thus, cannot make multi-layered matrix of binding partner obvious.

Additionally, as discussed in Section 7 of the Declaration, those skilled in the art would not have expected that such a multi-layered matrix would operate without "blocking" analyte access to the binding-partner molecules on the lower layers of the matrix. Contrary to the teachings and expectations of those skilled in the art, the present invention demonstrates the functionality and accessibility of the binding partner throughout the multi-layered three-dimensional matrix. As explained in the specification and in Section 7 of the Declaration, this result is achieved by using a photo-linking technique and by increasing the antibody concentration in the sorbent zones. Since the cited references teach significantly lower levels of the binding partner and do not teach a photo-linking technique, they do not make the multi-layered matrix of the present invention obvious.

In light of the foregoing, applicants respectfully submit that the '202 patent and the Immunoassay reference, either alone or in combination with other known techniques of the art, cannot make amended claims 1, 23 and 26 obvious. None of the cited references, either alone or in combination, would have motivated one skilled in the art to arrive at the present invention, which requires a substantial depletion of the analyte from the bulk solution and the concentration of the analyte on the microscopic sorbent zones comprising multi-layered matrix of analyte binding partner. Claims 2-4, 13-19, 24 and 25-28 depend, directly or indirectly, from claims 1, 23 and 26, and are not obvious for at least the same reasons. Accordingly, withdrawal of the rejection to claims 1-4, 13-19, 21 and 23-28 is respectfully requested.

Claims 1-4, 13-19, 21 and 23-28 are rejected under 35 U.S.C. § 103(a) as being unpatentable over the '202 patent in view of Ekins *et al.*, *Analytica Chimica Acta* (Analytica reference). This rejection is respectfully traversed.

The '202 patent and the state of the art prior to the present invention are discussed above. The Analytica reference cannot remedy the defect of the '202

patent. The Analytica reference discloses an ambient analyte immunoassay and does not teach or suggest a multi-layered matrix of analyte binding partner as required by amended claims 1, 23 and 26. Therefore, the '202 patent and the Analytica reference, either alone or in combination, cannot make claims 1, 23 and 26 obvious. Claims 2-4, 13-19, 24 and 25 depend, directly or indirectly, from claims 1 and 23, and are not obvious for at least the same reasons. Accordingly, withdrawal of the rejection to claims 1-4, 13-19, 21 and 23-28 is respectfully requested.

Claims 5-10 are rejected under 35 U.S.C. § 103(a) as being unpatentable over the '202 patent, and either the Immunoassay reference or the Analytica reference, in further view of Ullman *et al.* (U.S. Patent 5,512,659). Claim 11 is rejected under 35 U.S.C. § 103(a) as being unpatentable over the '202 patent, and either the Immunoassay reference or the Analytica reference in further view of Waggoner *et al.* (U.S. Patent 5,368,486). Claim 12 is rejected under 35 U.S.C. § 103(a) as being unpatentable over the '202 patent, and either the Immunoassay reference or the Analytica reference in view of Waggoner *et al.*, in further view of Lee *et al.* (U.S. Patent 5,453,505). Claim 20 is rejected under 35 U.S.C. § 103(a) as being unpatentable over the '202 patent, and either the Immunoassay reference or the Analytica reference in view of Northrup *et al.* (U.S. Patent 5,639,423). Applicants respectfully traverse these rejections.

As discussed above, the '202 patent, the Immunoassay reference, and the Analytica reference, either alone or in combination, cannot make amended claim 1 obvious, because they do not teach or suggest the binding assay of the present application, which requires the analyte to be substantially depleted from the sample and concentrated on microscopic sorbent zones comprising multi-layered matrix of analyte binding partner. Claims 5-10, 11, 12 and 20 depend, directly or indirectly, from claim 1 and cannot be made obvious by the '202 patent, the Immunoassay reference, and the Analytica reference for at least the same reasons.

Ullman *et al.*, Waggoner *et al.*, Lee *et al.*, and Northrup *et al.* cannot remedy the defect of the '202 patent, the Immunoassay reference, and the Analytica

reference, and are not relied upon by the Examiner for such. Ullman *et al.*, Waggoner *et al.*, Lee *et al.*, and Northrup *et al.* have no teaching whatsoever of a binding assay utilizing a plurality of sorbent zones containing an analyte-binding partner, let alone a binding assay which requires a multi-layered matrix of an analyte binding partner and a substantial depletion of analyte from the sample and its concentration in the sorbent zones. Therefore, none of the cited references, either alone or in combination, can motivate one skilled in the art to arrive at claims 5-10, 11, 12 and 20. Withdrawal of the rejection is, therefore, respectfully requested.

New claims 29-32 depend from claims 1, 23 and 26 and, thus, are patentable over the cited art for at least the same reasons as claims 1, 23 and 26. New independent claim 33 is patentable over the cited art because it requires microscopic sorbent zones comprising a multi-layer matrix of an analyte binding partner and the amount of the analyte binding partner immobilized in the sorbent zone with a diameter from 60 μm to 500 μm to be from 10^9 to 10^{12} molecules. New independent claim 34 is patentable over the cited art because it requires derivatizing a binding partner with a photolabile linker moiety to obtain a derivatized binding partner. New independent claim 34 is patentable over the cited art because it requires the analyte binding partner to be present in an amount from 10^9 to 10^{12} molecules per each sorbent zone with a diameter from 60 μm to 500 μm.

In view of the foregoing, it is respectfully submitted that the application is in condition for allowance. Reexamination and reconsideration of the application, as amended, are requested.

If for any reason the Examiner finds the application other than in condition for allowance, the Examiner is requested to call the undersigned attorney at the Los Angeles, California telephone number (213) 337-6700 to discuss the steps necessary for placing the application in condition for allowance.

If there are any fees due in connection with the filing of this response, please charge the fees to our Deposit Account No. 50-1314.

Respectfully submitted,
HOGAN & HARTSON L.L.P.

Date: November 6, 2002

By: _____
Wei-Ning Yang
Registration No. 38,690
Attorney for Applicant(s)

500 South Grand Avenue, Suite 1900
Los Angeles, California 90071
Phone: 213-337-6700
Fax: 213-337-6701

Version with markings to show changes made:

Please replace the text of claims 1, 23 and 26 with the following text:

1. (Twice Amended) A binding assay for sensing analyte mass in a liquid sample, comprising:

a) immobilizing an array on a substrate, wherein the array comprises a plurality of microscopic sorbent zones, wherein a microscopic sorbent zone comprises a multi-layer matrix of an analyte binding partner;

b) contacting a defined volume of sample believed to contain an analyte with at least one microscopic sorbent zone, the analyte binding partner in the microscopic sorbent zone being present in excess relative to the analyte, so that any analyte present in the defined volume is substantially depleted from the sample and concentrated on the microscopic sorbent zone to form an analyte capture complex with the analyte binding partner;

c) tagging the analyte capture complex with a fluorescent label;

d) illuminating the microscopic sorbent zone with a laser in the absence of liquid; and

e) detecting fluorescence emissions from any microscopic sorbent zone having an analyte capture complex tagged with a fluorescent label, thereby determining the analyte mass harvested from the defined volume of sample.

23. (Twice Amended) An analyte binding array for harvesting analyte from a liquid sample, the array comprising a plurality of microscopic sorbent zones immobilized on a substrate, wherein a microscopic sorbent zone comprises a multi-layer matrix of an analyte binding partner, the analyte binding partner being present in an amount sufficient to substantially deplete the analyte from a sample and concentrate the analyte on the microscopic sorbent zone, the microscopic zone being from about 60 to about 500 μm in diameter and the sample containing about 10^5 to about 10^{10} molecules of analyte per 100 μl of the sample, wherein a volume of the sample is from 20 to 500 μl.

26. (Twice Amended) A kit for use in a binding assay that senses analyte mass in a liquid sample of a defined volume, comprising an analyte binding array and a container comprising labeled binding partner,

wherein the analyte binding array comprises a plurality of microscopic sorbent zones immobilized on a substrate, wherein a microscopic sorbent zone comprises a multi-layer matrix of an analyte binding partner, the analyte binding partner being present in excess relative to the analyte, so that any analyte present in the defined volume of the sample is substantially depleted from the sample and concentrated on the microscopic sorbent zone to form an analyte capture complex with the analyte binding partner, and

the labeled binding partner having a fluorescent label and being capable of binding to an analyte bound by an analyte binding partner.